UNIVERSITÉ DU QUÉBEC À MONTRÉAL

UTILISATION DE DIFFÉRENTS DESCRIPTEURS D'ALIMENTATION POUR CARACTÉRISER LA COMMUNAUTÉ PARASITAIRE INTESTINAUX DU GOÉLAND À BEC CERCLÉ

MÉMOIRE

PRÉSENTÉ

COMME EXIGENCE PARTIELLE

DE LA MAÎTRISE EN BIOLOGIE

PAR

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NOVEMBRE 2013

Each species is a masterpiece,
a creation assembled with extreme care and genius.

Edward O. Wilson

ACKNOWLEDGEMENTS

This project would not have been possible without the contribution, involvement and support of my colleagues, family and friends. Each person mentioned hereafter as well as many others have all been instrumental in my accomplishments.

I would first like to thank my supervisor, Dr. Jonathan Verreault, as this project would not have been possible without his guidance, his many pep talks, his insightful perspectives on the subject, and the financial support he provided me through his NSERC grant. I thank him for taking me into his lab and giving me the opportunity to challenge myself in an unfamiliar subject. In the process, I gained invaluable skills and knowledge in a completely new field. He was always very well-organized and available to meet when I needed my focus to be brought back to what was essential. More importantly, I thank him for the delicious deserts he brought to all of our lab meetings.

I would like to give special thanks to three individuals who were essential to this project. I am greatly indebted to Dr. Dan McLaughlin (Concordia University) who took the time to teach me the meticulous and rigorous process of identifying avian helminths. I sincerely appreciate the many genuine talks we had and the wisdom he shared with me. I am grateful to Dr. Dave Marcogliese (Environment Canada) for his important contribution, the numerous article references he gave me, for his editing advice and for the many conversations we shared over a refreshing brewsky. I thank Dr. Sean Locke (Environment Canada) for his important collaborative role in this project. He trained me in the dissection of the intestinal tracts and in the identification process of the parasites, and he devoted much time to the statistical portion of this project.

I could not have completed this project without the help of my co-supervisor, Dr. Jean-François Giroux. He provided me with important ecological perspectives on ring-billed gulls, insights on my data analyses and help with the editing process of this thesis.

I thank Marie-Line Gentes for her part in organizing the field portion of the study and analyzing the GPS-tracking data. Thanks to Cynthia Franci, Sarah Marteinson and Stephanie Plourde Pellerin; sharing office space with them made this whole process a little more bearable. I would also like to mention my other lab mates: Élyse Caron-Beaudoin, Bernice Chabot-Giguère, Hubert Désilets, Angela Rose-Lapierre and John Forest. De plus, je tiens à donner un gros merci à nos deux techniciens de terrain Mathieu Tremblay et Francis St-Pierre, et à Laurine Bandet pour son assistance avec la dissection des tractus digestifs. Je voudrais aussi mentionner les membres du laboratoire GEOTOP-UQAM, Dr. Jean François Hélie et Mme. Agnieszka Adamowicz, pour leur aide dans les analyses d'isotopes stables.

I am grateful to the Canadian Society of Zoologists (CSZ) and TOXEN for the travel grants they provided me and for the opportunity they gave me to present at the 2012 and 2013 CSZ conferences.

Last but certainly not least, I am forever grateful to my loving parents, my four incredible siblings and to the rest of my family. I would like to send a particular shout out to my best friend Chris: I thank her for being my rock. Finally, I would like to show my appreciation for all the cherished friendships I have made along the years.

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List of Abbreviations and Symbols

ANOSIM	Analysis of Similarity
C	Carbon
g	Gram
GI	Gastrointestinal
GPS	Global Positioning System
km/h	Kilometers per hour
m	Meter
mg	Milligram
mL	Milliliter
N	Nitrogen
PC	Principal Component
SIMPER	Similarity Percentage
SD	Standard Deviation
°C	Degree Celsius
$\delta^{13}C$	Carbon Stable Isotope
$\delta^{15}N$	Nitrogen Stable Isotope

RÉSUMÉ

Le régime alimentaire d'une espèce a un rôle important dans la structure d'une communauté parasitaire intestinale au point de vue de la diversité et de l'abondance des différents parasites. Il existe cependant des variations intraspécifiques parfois importantes dans les communautés parasitaires chez une même espèce qui peuvent être expliquées par des variations dans leur diète. De plus, plusieurs espèces aviaires profitent des milieux anthropiques pour se nourrir. Ainsi, des altérations aux taux d'interactions hôtes-parasites peuvent engendrer des changements à la structure des communautés parasitaires. Cette hypothèse a rarement été étudiée dans un modèle aviaire sauvage. Le but de cette recherche était d'investiguer les variations dans les communautés parasitaires intestinales d'une espèce aviaire urbaine et omnivore, le goéland à bec cerclé (*Larus delawarensis*) en période de nidification (n=34) sur l'Ile DesLauriers, et d'étudier si une diète plus axée sur la nourriture d'origine anthropique se traduit en une charge parasitaire totale plus basse. En 2011, trois méthodes complémentaires ayant des échelles temporelles différentes furent utilisées pour caractériser leur régime alimentaire. Les contenus stomacaux furent récoltés donnant un aperçu de leur diète pendant les dernières 24 heures. Pour la première fois dans une étude parasitaire, le lien entre les aires d'alimentation récemment visitées et la communauté parasitaire d'une espèce aviaire fut étudié en utilisant des unités GPS miniatures qui localisaient les déplacements des oiseaux pendant 30 à 72 heures. Des analyses d'isotopes stables d'azote ($\delta^{15}N$) et de carbone ($\delta^{13}C$) (foie) furent faites donnant des renseignements sur l'assimilation de nourriture dans les derniers 10 jours. Parasites les plus communs furent *Wardium* spp., *Diplostomum* spp. et *Eucoleus* spp., transmit par les crustacés, les poissons d'eau douce, et les vers de terre, respectivement. Selon les données GPS, la proportion de temps passé dans les différentes aires d'alimentation par les goélands à bec cerclé tendait à être associée à la structure des communautés parasitaires ($p = 0.07$). Cependant, les variations dans les communautés parasitaires intestinales étaient mieux expliquées par les signatures d'isotopes stables d'azote et de carbone ($p < 0.001$). D'ailleurs, le ratio $\delta^{15}N$ était significativement plus bas chez les individus ayant une abondance totale de parasites moindre ($p = 0.03$). Il a récemment été établi que la nourriture anthropique a une signature ^{15}N appauvri. Les résultats de la présente étude suggère donc que les individus se nourrissant plus de nourriture anthropique sont moins infestés de parasites. Ces résultats semblent démontrer que la disponibilité de nourriture d'origine anthropique peut avoir un effet sur la structure des communautés parasitaires d'une espèce aviaire nichant en milieu urbain. Ceci pourrait avoir des implications sur le succès reproducteur du goéland à bec cerclé.

Mots clés : Télémétrie, oiseaux, Laridae, régime alimentaire, isotopes stables, urbanisation

ABSTRACT

The diet of a host has an essential role in shaping the structure of intestinal parasite communities, in terms of abundance and diversity of the different parasites. Parasite community composition is often highly variable among birds of the same species. A number of avian species breeding near urbanized areas may benefit from anthropogenic sources of food. As a result, a shift away from natural (terrestrial and more importantly aquatic) towards non-parasitized anthropogenic food sources may possibly have an impact on the structure of endoparasite communities. In order to investigate the link between parasite community structure and diet, in 2011, we examined the abundance of intestinal parasites and characterized the feeding ecology of an omnivorous urban species breeding on Deslauriers Island, the ring-billed gull (*Larus delawarensis*) (n = 34), using three methods with distinct time scales. Stomach contents were analysed providing information on diet over the last 24 hours. For the first time in a parasitological study, miniature GPS-based dataloggers were used to monitor foraging habitat use strategies during the previous 30 to 72 hours. The locations of the birds corresponded to specific foraging areas (anthropogenic, agricultural, freshwater areas and wastewater treatment basins) associated with different trophically-transmitted parasites. Liver carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) stable isotopes were determined offering insight into food assimilation during the last week. The three most abundant and prevalent parasites found in ring-billed gulls were *Wardium* spp., *Diplostomum* spp. and *Eucoleus* sp., transmitted from crustacean, freshwater fish and earthworm intermediate hosts, respectively. No relationship was found between stomach contents and parasites. Foraging habitat use strategies, obtained from the GPS-tracking devices, had a tendency to be associated with parasite community structure ($p = 0.07$). However, intraspecific variations in parasite communities were best explained by $\delta^{13}C$ and $\delta^{15}N$ ratios ($p < 0.001$). In fact, $\delta^{15}N$ ratios were lower in individuals with poorer parasite infection levels ($p = 0.03$). It has previously been shown that anthropogenic foods are depleted in ^{15}N. Thus, the results from this study suggest that individuals feeding lower in the food chain or similarly feeding more on anthropogenic food sources are reducing their encounter with trophically-transmitted parasites. This demonstrates that the availability of anthropogenic food sources may be having an impact on local parasite community structures and on the reproductive success of this widespread urban species.

Keywords: Geolocation, birds, Laridae, foraging ecology, stable isotopes, urbanization

INTRODUCTION

The study of communities and the determinant factors shaping their structure and composition has always been an important element of ecology. However, the scope of these studies is usually focused on the community structure of wildlife ecosystems which include higher taxa such as amphibians, reptiles, birds and mammals. Smaller organisms such as parasites are often overlooked, probably as a result of their size and the logistical challenges associated with studying them. Yet, parasites most likely outnumber free-living species and nearly all free-living organisms are host to parasites. In fact, if their abundance is high enough, parasites may be involved in regulating host populations. Therefore, understanding the various factors which may influence parasite populations can provide valuable information in ecological studies.

Gastrointestinal parasites within a host are those which result from the ingestion of infected prey. The interactions between predators and their prey, like in omnivorous species, is probably the most important proximate factor shaping gastrointestinal parasite communities. However, studies which have attempted to demonstrate this have often been unable to successfully establish the link between prey diet and parasite communities. Furthermore, most previous studies have only used stomach contents as an ecological indicator of host diet. More recently, researchers have been able to incorporate additional information on diet by using carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) stable isotope analyses of different tissues. ^{13}C stable isotope provides information on the sources of carbon in a food web whereas ^{15}N stable isotope provides information on the trophic level of a host within the food web. Still, studies that have used both these methods lack precise information on foraging locations used by a host. Where a host feeds will directly influence the kinds of prey it encounters (or not). There is a direct or indirect association between habitat use strategies and parasite community composition. Recent technological advances in GPS (global positioning system)-based tracking devices have made it possible to determine the movements of a variety of small- to medium-size species during a particular time-frame.

The ring-billed gull (*Larus delawarensis*) is an omnivorous species often breeding near urban areas and displaying intraspecific variations in feeding behaviour. Herein, this

model species is used to evaluate how these intraspecific variations in diet may cause intraspecific variations in gastrointestinal parasite communities. The importance of diet as a factor influencing parasite community structure is evaluated here. In 2011, three different dietary descriptors with distinct time-frames were used to characterize the diet of ring-billed gulls breeding on Ile Deslauriers (n = 34), a colony situated in the St. Lawrence River in the Greater Montreal Region. Stomach contents were sampled providing information on very recent food consumption (<24 hrs). Miniature GPS data-loggers were attached to the birds (tail feathers) for a period of two to three days recording tracking information on their habitat use strategies during that time-frame. Liver $\delta^{13}C$ and $\delta^{15}N$ were obtained offering insight into food assimilation during the last week or so. To date, no study has been done on the relationship between habitat use strategies of a host and its parasite community, making this the first study to investigate how the utilization of specific foraging areas, associated with particular prey, influence host-parasite interactions. Most other parasitological studies focus on one parasite taxon (e.g. nematodes), or sometimes use only presence/absence data or other collapsed versions of the parasite community (e.g. subsamples of the entire infracommunity or diversity indices); this study stands apart from others in that the entire parasite infracommunity of each host sampled was included in the analyses.

The findings of this Master's research project are reported in the form of an article-based thesis in the present document comprising of two chapters. The first chapter reviews the current literature pertinent to the global comprehension of this study. These subjects include general background information on parasite life cycles and terminology, as well as a review of factors involved in shaping their communities along with the research that has previously been done on this topic. Finally, a brief biological account of the model organism is provided and the specific objectives of the study are then stated. The second chapter, written in the form of a manuscript intended for publication in a scientific journal, is an in depth analysis of the relationship between the feeding behaviour of ring-billed gulls, as described by the three dietary indicators used in this study, and their gastrointestinal parasite community assemblages.

CHAPTER I

LITERATURE REVIEW

1.1 Parasite life cycles

Parasites can be dichotomously classified into two large groups based on their interactions with the host: ectoparasites and endoparasites. Parasites that live on the surface of the host are termed ectoparasites and include many arthropods and monogeneans. Endoparasites, on the other hand, live in organs or tissues within the host. They comprise of unicellular organisms called microparasites which include protozoans, viruses and bacteria as well as multicellular organisms such as myxozoans (Marcogliese, 2004). Infection by these parasites often results in disease. In contrast, macroparasites are multicellular organisms and include trematodes, cestodes, nematodes, acanthocephalans among others, whose effects on the host are often sublethal and therefore, less obvious. However, simultaneous multiple co-infections by several parasites may occasionally have severe compounding effects resulting in the death of the host (parasite-induced host mortality). Transmission may involve a direct life cycle, requiring only one host or an indirect life cycle, with one or more free-living infective stages which are either passively (via ingestion) or actively (via penetration) transmitted to one or more obligate intermediate hosts. In the intermediate host, the parasite undergoes some developmental and morphological change but does not reach maturity. The infected intermediate host is then ingested by the "definitive host" where it reaches sexual maturity in the gastrointestinal (GI) tract (Bush *et al.*, 2001). The time required to reach sexual maturity depends both on the type of parasite considered and the conditions found within the host. Most macroparasites have indirect life cycles, except monogeneans and some nematodes which have direct life cycles (Bush *et al.*, 2001). Occasionally, parasites infect "paratenic" hosts, in which no development occurs and which are not essential in the parasite's life cycle. Instead, paratenic hosts serve as a link between hosts that may be ecologically or trophically separated.

1.2 Parasite communities

In ecology, the concept of community is defined as an assemblage of more than one organism living together in a particular spatio-temporal unit (Bush *et al.*, 1997). Based on this definition, the community of parasites living within a single host is termed

"infracommunity" whereas a "component community" refers to all parasites associated with a subsample of a particular host species (Bush *et al.*, 1997). Abundance and prevalence are terms used to describe parasite communities where abundance denotes the number of individuals of a particular parasite within a single host; mean abundance is the number of individuals of a particular parasite within the component community; and prevalence is calculated as the number of hosts infected by a particular parasite species (Bush *et al.*, 1997).

1.2.1. Factors shaping parasite communities

A variety of factors influence the abundance and prevalence of gastrointestinal parasites of a host, i.e. the infracommunity of the host. The mere presence of a parasite in a host is considered to be the result of historical evolutionary processes both at the host level and at the parasite level (Janovy, Clopton and Percival, 1992; Poulin, 1995). For instance, the age of the host lineage has been suggested to affect species richness in the way that older host groups have a longer period of time to accumulate parasite species (Gregory, Keymer and Harvey, 1995). In order for a new parasite to successfully establish itself in a host, the chemical and physical properties of the host environment must be suitable for its survival; and the host ecology must be compatible with the life cycle of the parasite (Janovy, Clopton and Percival, 1992; Vickery and Poulin, 1998). Throughout evolutionary time, parasites have evolved concurrently with their host. Immediate factors that control host-parasite interactions and encounters are those which include ecological properties of the host and to a lesser extent, the parasite (Holmes and Price, 1980; Janovy, Clopton and Percival, 1992). For instance, it has been found that the similarity of parasite component communities decreases with the geographic range of the host. The influence of geographic range and vagility of a host have been extensively studied across multiple taxa including mammals (e.g. Poulin, 2003) and fishes (e.g. Poulin and Morand, 1999) and more recently, in an avian species, the ring-billed gull (*Larus delawarensis*) (Locke *et al.*, 2011). In the latter study, the authors also found that host age class influenced parasite communities (Locke *et al.*, 2011). The authors compared the mean abundance of parasites between chicks (fed by parents), juvenile of the year (feeding on their own) and adult ring-billed gulls, and found increasing diversity of parasites in older gulls. Another host characteristic recognized as an important predictor of

parasite communities is body size (often measured as an index of structural size, body length or mass). Many researchers apply island biogeography theory to hosts: the hosts are analogous to an island where the availability of resources is directly associated with its area. Accordingly, larger hosts are expected to have a higher abundance and species richness of parasites because they may provide a greater number of available niches and because larger individuals tend to eat more (i.e. ingest more infected intermediate hosts) (Gregory, Keymer and Harvey 1991; Sasal, Niquil and Bartoli, 1999). Immigration and extinctions of parasites within a host are limited by the amount of resources available (i.e. host body size) (Kuris, Blaustein and Alio, 1980; Vickery and Poulin, 1998). Furthermore, due to inter- and intraspecific competition between parasites for food and space, there is a threshold for the number of parasites that can coexist within the same habitat (i.e. the host) (Sasal, Niquil and Bartoli, 1999; Lagrue and Poulin, 2008). In many vertebrate species, differences in parasitism have also been associated with gender. Generally, in species of mammals exhibiting sexual size dimorphism, the larger sex (male or female) shows biases to parasitism (Moore and Wilson, 2002). These gender-specific differences have been attributed, for example, to differential investment of energy into growth rather than immunity by males. This also results in sexual size dimorphism (Moore and Wilson, 2002).

Under natural conditions, host diet is arguably the most important factor affecting gastrointestinal parasite communities. Variations in prey feeding behaviour and consequently variability in intermediate hosts ingested by a predator, likely contribute to variations in their parasite community composition (Knudsen, Curtis and Kistoffersen, 2004). As such, many authors strongly advocate the use of parasites within food web studies because intestinal parasites are transmitted through predator-prey interactions (Knudsen, Klemetsen and Staldvik, 1996; Lafferty *et al.*, 2008; Valtonen, Marcogliese and Julkunen, 2010); while other researchers have attempted to understand the role of host diet on parasite community composition. This subject has been studied in fishes (e.g. Johnson, Hesslein and Dick, 2004; Knudsen, Curtis and Kistoffersen, 2004; Munoz, Grutter and Cribb, 2006; Locke *et al.*, 2013), some mammal species (e.g. Sinisalo *et al.*, 2006; Kirkova, Raychev and Georgieva, 2011) and in birds (e.g. Robinson, Forbes and Hebert, 2009). The authors of these studies have used stomach contents and/or carbon and nitrogen stable isotopes as descriptors of host diet and feeding ecology in order to investigate this.

1.3 Descriptors of host diet used in parasite community studies

1.3.1 Stomach contents

Over the years, a number of methods have been developed to investigate the diet of wild species to understand parasite communities. A conventional technique has been the analysis of stomach contents (Hyslop, 1980) which provide information on very recent (less than 24 hrs) feeding behaviours. For instance, Knudsen, Curtis and Kistoffersen (2004) evaluated the degree of feeding specialization on littoral vs. pelagic crustaceans of Arctic char (*Salvelinus alpinus*) from a lake in northern Norway by using stomach contents in relation to helminths transmitted by pelagic copepods and those transmitted by benthic amphipods. A correlation between the prey items found in the gut contents and the intestinal parasites indicated segregation between two feeding strategies, benthivory and planktivory, within members of the same char population (Knudsen, Curtis and Kistoffersen 2004). In another study done on five Great Barrier Reef fish species from the Labridae family, cluster analyses of the relatedness of fish species according to their parasites and dietary items collected were both highly correlated together (Fig. 1.1).

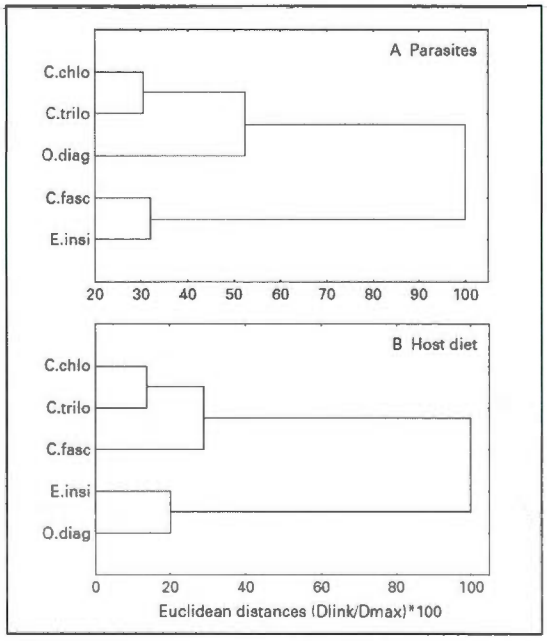

Figure 1.1 (A) Cluster analysis based on the presence/absence of all parasites and **(B)** cluster analysis for diet categories of five Labridae fishes from the Great Barrier Reef (from Munoz, Grutter and Cribb, 2006)

1.3.2 Stable isotopes

Stable isotopes of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) have been used in trophic web ecology in both terrestrial and aquatic ecosystems since the late 1970's. Since then, technological advances have made their application in wildlife studies increasingly easy. Organs and tissues (e.g. muscle, liver, feathers, etc.) incorporate nutrients from diet differentially, and therefore can provide information on feeding habits during varying timescales. For instance, liver $\delta^{13}C$ and $\delta^{15}N$ profiles in birds reflect diet assimilation over a period of about a week, whereas plasma $\delta^{13}C$ and $\delta^{15}N$ reflect only a few days' worth of information on food consumption (Hobson and Clark, 1992). The trophic position of an organism may directly be linked to its parasite infection level. It has been shown that predators feeding higher up in the food chain have higher $\delta^{15}N$ values and are likely ingesting more parasites (Robinson, Forbes and Hebert, 2009). $\delta^{13}C$ is useful for tracking different carbon sources in food chains. For example, in terrestrial ecosystems, $\delta^{13}C$ values distinguish C_3 (e.g. wheat) from C_4 (e.g. maize) based food chains because C_3 primary consumers are depleted in ^{13}C whereas C_4 plants are enriched in it (Kelly, 2000). Moreover, in aquatic ecosystems, $\delta^{13}C$ can be used to differentiate between ^{13}C enriched littoral (benthic periphyton) versus ^{13}C depleted pelagic (phytoplankton) sources (France, 1995). Given that phytoplankton and periphyton are intermediate hosts to different parasite species, an association between $\delta^{13}C$ and parasite communities may be inferred. In a study done on double-crested cormorants (*Phalacrocorax auritus*) from Lake Ontario, the authors used muscle $\delta^{13}C$ and $\delta^{15}N$ values and related them to the abundance of *Contracaecum* spp. (Robinson, Forbes and Hebert, 2009), nematodes with an indirect life cycle requiring crustaceans as its first intermediate host and fish as its second intermediate host (Anderson, 2000). Robinson, Forbes and Hebert (2009) found a significant negative correlation between the abundance of *Contracaecum* spp. and $\delta^{13}C$, but no relationship with $\delta^{15}N$ which they suggest means that *Contracaecum* spp. are not necessarily aggregated in larger prey, higher in the food chain, but instead are more present in pelagic areas.

Stable isotopes offer much more comprehensive information regarding the feeding ecology of an organism than stomach contents. Still, there are limitations to this method,

especially pertaining to its application in feeding ecology studies of omnivorous species such as the ring-billed gull. Ring-billed gulls forage from both terrestrial and aquatic ecosystems which complicates the interpretation of its stable isotope signatures. They also take advantage of anthropogenic sources of food (e.g. fast food) with highly contrasting $\delta^{13}C$ and $\delta^{15}N$ profiles compared to natural foods. For instance, Jahren and Kraft (2008) found that $\delta^{13}C$ values in fast food products such as beef, chicken and fries were highly variable. ^{13}C content depended on the origin of the fast food (McDonald vs Wendies vs Burger King) as well as the fast food product. ^{15}N composition was poor, from as low as 1.5 in McDonald's chicken (Jarhen and Kraft, 2008). The exploitation of anthropogenic foods by ring-billed gulls likely confounds their stable isotope signatures further (see section 1.4.2 for a detailed description of the feeding ecology of ring-billed gulls).

1.3.3 GPS-based telemetry

In order to fill important information gaps related to habitat use strategies left from stable isotope and stomach content analyses, monitoring of foraging habitat use strategies can be obtained from miniature bird-borne GPS (global positioning system) tracking units. With time, GPS loggers have become increasingly small (<20 g) allowing for the study of movements of a variety of small- to medium size birds (Burger and Shaffer, 2008) without seemingly affecting their flight patterns. The only observed behavioural changes appear to occur immediately after handling the bird to affix the devices (Weimerskirch *et al.*, 2005). Depending on the battery duration and programing script of the data-loggers, these devices record the locations of the bird at specific time intervals during a given time-frame. After processing the data revealed from these recordings, information on time spent in a particular habitat, speed and altitude can be obtained. From these data, foraging areas visited can be documented and characterized with a great degree of precision (5-10 m), and thus intraspecific variations in behavioural feeding patterns may be inferred. For instance, a telemetry-based study on black-browed albatrosses (*Thalassarche melanophrys*) from South Georgia revealed seasonal sexual segregation in foraging habitat use (Phillips *et al.*, 2004). In this study, female black-browed albatross foraged northward farther away from the colony and for longer periods of time whereas males foraged more eastward during shorter feeding

trips. In contrast, there was no sexual segregation in foraging areas during the chick-rearing period (Phillips *et al.*, 2004). There still remain some limitations in the use of GPS data loggers (e.g. duration of battery, the necessity of recapturing individuals to retrieve the units) (Burger and Shaffer, 2008); therefore, combining *multiple* dietary descriptors in feeding ecology studies can provide a much more complete idea of the foraging behaviour of a particular species.

1.4 The ring-billed gull as a model species

There are several reasons why ring-billed gulls are an interesting model organism to address the present research objectives. In general, it was important to choose a species with an otherwise stable population so that the removal terminal samples would not have an impact on their total population count. The ring-billed gull is widespread, abundant and is currently listed as a species of "least concern" (IUCN, 2011). According to Canadian Wildlife Service surveys, it is the most widespread larid in southern Quebec with its population tripling between 1979 and 1994, from 36,000 to 125,000 breeding pairs (CWS, 2013). The increase in their population over the past decades has often been attributed to a higher availability of anthropogenic food sources (Brousseau, Lefebvre and Giroux, 1996; Belant, Ickes and Seamans, 1998). Since then, their population has seen a slight decline (J.-F. Giroux, personal communication, 2013). More importantly, however, ring-billed gulls make an ideal model organism for this study because intraspecific variations in their feeding behaviour (see section 1.4.2 below) may be used to investigate the influence that diet has on shaping parasite communities (Locke *et al.*, 2011).

1.4.1 Intraspecific variations in the feeding ecology of ring-billed gulls

The feeding ecology of ring-billed gulls has been extensively studied across North America (Haymes and Blokpoel, 1978; Welham, 1987; Burger, 1988; Belant, Ickes and Seamans, 1998) and especially in the St. Lawrence River and Great Lakes Region (Jarvis and Southern, 1976; Lagrenade and Mousseau, 1981; Brousseau, Lefebvre and Giroux, 1996; Brown and Ewins, 1996; Patenaude-Monette, 2011). As omnivores, ring-billed gulls feed on

a variety of food items including fish, molluscs, crustaceans, earthworms, terrestrial and aquatic adult and larval insects, snails, small mammals and birds, plants; and exploit human-related sources of food such as grains from agricultural fields, fast foods, and human refuse in cities and landfills (Brousseau, Lefebvre and Giroux, 1996; Belant, Ickes and Seamans, 1998). Given their omnivorous feeding behaviour, anthropogenic and natural food availability influence their dietary habits (Haymes and Blokpoel, 1978), and therefore may influence their gastrointestinal parasite infracommunities. Their feeding behaviour has also been found to vary with the season due to insect emergences or nearby agricultural practices (Brousseau, Lefebvre and Giroux, 1996).

1.4.2 The importance of ring-billed gulls in the transmission of local parasites

Despite the large number of studies which have been done on the ecology of ring-billed gulls, there remain few studies on their parasite communities and the factors which shape them. To date, there are only two published papers (Vermeer, 1969; Locke *et al.*, 2011) and one Master's thesis (Levy, 1997) on the parasite assemblages of this species (Table 1.1). The high abundance of these birds is likely having an impact on the rate of transmission of their parasites which may have implications for the parasite communities in the region (Marcogliese *et al.*, 2001). In a study where spottail shiners (*Notropis hudsonius*) were surveyed along the St. Lawrence River for an eyefluke (*Diplostomum* spp.) which affects the lens of the fish's eyes, it was found that the mean abundance of *Diplostomum* spp. were highest near large ring-billed gull colonies (Marcogliese *et al.*, 2001). Hence, a comprehensive study on the factors shaping gastrointestinal endoparasite communities of such an abundant avian species breeding in the Greater Montreal Region would contribute to this area of research.

Table 1.1

Parasite genera (present/absent) recovered from ring-billed gulls (*Larus delawarensis*) from Miquelon Lake, AB (Vermeer, 1969) and along the St. Lawrence River, QC (Levy, 1997)

	Vermeer, 1969	Levy, 1997
Cestodes		
Wardium spp.		✓
Choanotaenia sp.		✓
Microsomacanthus sp.		✓
Anomotaenia sp.		✓
Drepanidotaenia sp.		✓
Tetrabothrius sp.		✓
Aploparaxis sp.		✓
Ophryocotyle sp.		✓
Hymenolepis sp.	✓	
Paricterotaenia sp.	✓	
Diphyllobothrium spp.	✓	✓
Lateriporus sp.	✓	
Schistocephalus sp.	✓	
Trematodes		
Plagiorchis spp.	✓	✓
Diplostomum spp.	✓	✓
Stephanoprora sp.		✓
Apophallus sp.		✓
Mesoophorodiplostomum sp.		✓
Echinostoma spp.	✓	✓
Cotylyrus sp.	✓	✓
Maritreminoides sp.		✓
Cardiocephalus sp.		✓
Himasthala sp.		✓
Echinoparyphium sp.	✓	
Austrobilharzia sp.	✓	
Nematodes		
Cosmocephalus sp.		✓
Tetrameres sp.		✓
Eucoleus spp.		✓
Aprocta sp.	✓	

1.5 Hypotheses and objectives

The diet of a host is known to be an important factor influencing parasite communities. Within an omnivorous species such as the ring-billed gull, variations in feeding behaviour likely have an impact on the structure and composition of their gastrointestinal parasite communities. Furthermore, the incorporation of anthropogenic sources of food in their diet may be reducing their encounter with trophically-transmitted parasites.

The general objective of this study is to investigate variations in the parasite infracommunity structure of ring-billed gulls breeding in the St. Lawrence River, in the Greater Montreal Region by studying their feeding behaviour using three dietary descriptors with distinct time-frames (Fig. 1.2): stomach contents, liver $\delta^{13}C$ and $\delta^{15}N$ profiles and foraging habitat use information.

Hypothesis 1: Intraspecific variations in the feeding behaviour of ring-billed gulls are associated with variations in their parasite community structure.

Objective 1: To test whether the gastrointestinal parasite communities of ring-billed gulls breeding in the St. Lawrence River can be explained using three different dietary descriptors covering distinct time-frames (Fig 2.1); and whether birds with similar feeding behaviours have similar parasite communities.

Hypothesis 2: Ring-billed gulls exploiting anthropogenic sources of food reduce their encounter with trophically-transmitted parasites and as a result, should have a lower total abundance of gastrointestinal helminths.

Objective 2: To test whether ring-billed gulls relying less on natural food webs and more on anthropogenic-related foods have a lower abundance and richness of trophically-transmitted gastrointestinal parasites.

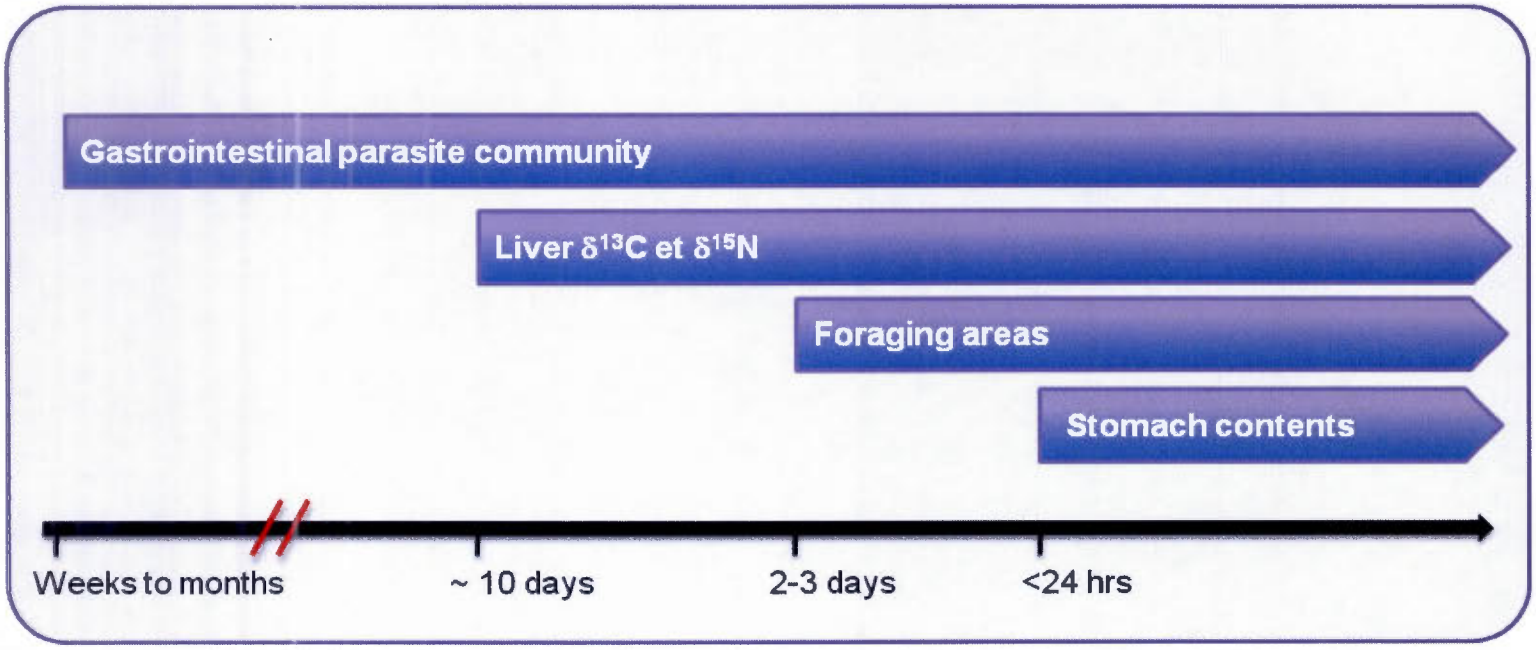

Figure 1.2 Timeline showing the time-frame associated with the different diet descriptors used in this study

CHAPTER II

EXPLAINING THE INTESTINAL PARASITE COMMUNITY OF NESTING RING-BILLED GULLS USING MULTIPLE DIETARY DESCRIPTORS

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2.1 Résumé

Il existe des variations intraspécifiques parfois importantes dans les communautés parasitaires chez une même espèce qui peuvent être expliquées par des variations dans leur diète. Plusieurs espèces aviaires profitent des milieux anthropiques pour se nourrir. Ainsi, des altérations aux taux d'interactions hôtes-parasites peuvent engendrer des changements à la structure des communautés parasitaires. Cette hypothèse a rarement été étudiée dans un modèle aviaire sauvage. Le but de cette recherche était d'investiguer les variations dans les communautés parasitaires intestinales d'une espèce aviaire urbaine et omnivore, le goéland à bec cerclé (*Larus delawarensis*) nichant dans la région de Montréal. Trois méthodes complémentaires ayant des échelles temporelles différentes furent utilisées pour caractériser leur régime alimentaire. Les contenus stomacaux furent récoltés donnant un aperçu de leur diète pendant les dernières heures. Le lien entre les aires d'alimentation récemment visitées et la communauté parasitaire d'une espèce aviaire fut étudié en utilisant des unités GPS miniatures qui localisaient les déplacements des oiseaux pendant deux à trois jours. Des analyses d'isotopes stables d'azote ($\delta^{15}N$) et de carbone ($\delta^{13}C$) (foie) furent faites donnant des renseignements sur l'assimilation de nourriture dans les derniers dix jours. Aucun lien n'a été établi entre les contenus stomacaux et les communautés parasitaires. Selon les données GPS, la proportion de temps passé dans les différentes aires d'alimentation par les goélands à bec cerclé tendait à être associée à la structure des communautés parasitaires Cependant, les variations dans les communautés parasitaires intestinales étaient mieux expliquées par les signatures d'isotopes stables d'azote et de carbone. D'ailleurs, le ratio $\delta^{15}N$ était significativement plus bas chez les individus ayant une abondance totale de parasites moindre ce qui suggère que les individus se nourrissant plus bas dans la chaîne alimentaire ou de nourriture anthropique sont moins infestés de parasites. Ces résultats semblent démontrer que la disponibilité de nourriture d'origine anthropique peut avoir un effet sur la structure des communautés parasitaires d'une espèce aviaire nichant en milieu urbain.

Mots clés: Parasites, télémétrie, isotopes stables, comportement de quête alimentaire, urbanisation, Laridae, Fleuve Saint-Laurent

2.2 Abstract

Parasite communities are often highly variable among birds of the same species. This can largely be explained by intraspecific variations in feeding habits. Furthermore, many avian species breeding near cities profit from anthropogenic sources of food. As a result, a shift away from natural (terrestrial and mainly aquatic) towards non-parasitized anthropogenic food sources is possibly having an impact on the structure of endoparasite communities. Few studies have investigated this. In order to study the link between parasite community structure and diet, we examined intestinal parasites and characterized the feeding ecology of an omnivorous species breeding in the Greater Montreal Region, the ring-billed gull (*Larus delawarensis*), using three methods with distinct time scales. Stomach contents were analysed providing information on diet over the last few hours. Miniature GPS-based dataloggers were used to monitor foraging habitats associated with different trophically-transmitted parasites during the previous two to three days. Liver carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) stable isotopes were determined offering insight into food assimilation during the last ten days. No relationship was found between stomach contents and parasites. Foraging habitat use strategies, obtained from the GPS-tracking devices, had a tendency to be related with parasite community structure. However, intraspecific variations in parasite communities were best explained with $\delta^{13}C$ and $\delta^{15}N$ ratios. In fact, $\delta^{15}N$ ratios were inferior in individuals with poorer parasite infection levels which suggests that individuals feeding lower in the food chain or similarly feeding more on anthropogenic food sources are reducing their encounter with trophically-transmitted parasites. These results demonstrate that the availability of anthropogenic food sources may be having an impact on parasite community structures on local hosts adapted to human-dominated habitats.

Keywords: Parasites, geolocation, stable isotopes, foraging ecology, urbanization, Laridae, St. Lawrence River

2.3 Introduction

As urbanization continues to encroach on wildlife habitat, bird species which are able to adapt to these landscape changes often thrive by taking advantage of anthropogenic sources of food. Individuals relying less on natural food webs and more on human-made foods are likely reducing their encounter with parasite infected prey. This shift away from natural food is likely having an impact on their parasite communities (Delgado and French, 2012). To our knowledge, the only study to investigate the effect of urbanization on parasite abundance was done on red foxes (*Vulpus vulpus*). The authors found that in urban areas, this species showed shifts in dietary behavior and encountered fewer intermediate hosts, resulting in lower prevalence of *Echinococcus multilocularis* than rural foxes (Fischer *et al.*, 2005). Host-parasite interactions in avian species occupying and/or breeding in urbanized areas are poorly understood (Delgado and French, 2012). Yet avian hosts make particularly interesting models to study the importance of urbanization on the diet, and also parasites, especially highly vagile and omnivorous birds which forage in both aquatic and terrestrial habitats. Such hosts necessarily acquire different species of parasites from the two ecosystems. However, these same reasons make studying the diet of birds inherently difficult. Thus, most research on the link between diet and parasite communities has focused on fish hosts (e.g. Sasal, Niquil and Bartoli, 1999; Munoz, Grutter and Cribb, 2006; Locke *et al.*, 2013).

Ring-billed gulls (*Larus delawarensis*) are an urban-adapted omnivorous species which forage in both aquatic and terrestrial ecosystems. The most recent survey of their gastrointestinal parasites (Locke *et al.*, 2011) showed that while most of the helminths recovered were transmitted through aquatic hosts (fish and arthropods), some had terrestrial life cycles. Ring-billed gulls breeding in the Greater Montreal region, a heterogeneous area characterized by freshwater habitat, agricultural fields (soybean, maize and cereals), urban zones and landfills (between 8-63 km from the colony), make particularly interesting populations for examining the influence of urbanization on diet and on gastrointestinal parasite communities. The increase in human populations and the increased availability of agricultural habitats in this region have been considered the main contributing factors to the success of this species in North America. Ring-billed gulls have a tendency to take advantage of human refuse and agricultural lands as sources of food (Jarvis and Southern, 1976;

Welham, 1987), as seen in other gull species (e.g. yellow-legged gull, *L. michahellis*) (Duhem *et al.*, 2008). Brousseau, Lefebvre and Giroux (1996) found that the proportion of human refuse in regurgitations of ring billed-gull chicks increased markedly between 1978 and 1993. Concurrently, between the 1970's and 1990's, their North American population showed a remarkable increase, especially near urban areas (Canadian Wildlife Service, unpubl. data). An increased dependency on non-natural food items may be reducing the encounter between ring-billed gulls and infected prey, which in turn may be affecting their gastrointestinal parasite communities.

Several methods have been used to investigate the diet of birds. Stomach content analysis, a conventional method applied widely in feeding ecology studies (Hyslop, 1980), provide very short term (only a few hours) information on an animal's diet unless they are collected over a long period of time (Inger and Bearhop, 2008). Furthermore, the variability in digestibility of food items often makes stomach content characterization biased against fast digesting prey like amphipods or cnidarians vs. slow digesting foods such as vegetation or seeds (Jackson, Duffy and Jenkins, 1987). In the last 25 years, carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) stable isotopes increasingly have been integrated in foraging ecology studies (Kelly, 2000; Inger and Bearhop, 2008) and since have been used in parasitological studies to gain better insight into food web structures (e.g. Robinson, Forbes and Hebert, 2009). Stable isotopes offer a more comprehensive picture of the feeding behavior of a species than stomach contents; and depending on the tissue sampled, stable isotopes can integrate dietary information over variable temporal scales. Still, they cannot provide information regarding specific habitat use strategies. Recently, Caron-Beaudoin *et al.* (submitted) used global positioning system (GPS)-tracking information to determine foraging habitat use strategies of ring-billed gulls breeding on an island in the St. Lawrence River. The authors also related GPS-tracking information to $\delta^{13}C$ and $\delta^{15}N$ ratios of this urban-adapted gull species. Blood $\delta^{15}N$ revealed that individuals foraging predominantly in anthropogenic-related areas had significantly lower $\delta^{15}N$ values; and it was also found that individuals tended to conserve particular foraging patterns throughout the incubation period, exhibiting a tendency to be "local specialists". This also has been observed for a different ring-billed gull population breeding in Dog Lake, Alberta with access to agricultural lands, freshwater and dump sites

(Welham, 1987). In the study, the authors found that ring-billed gulls of Dog Lake forage mainly in agricultural areas.

The objective of the present study was to evaluate the effect of urbanization on gastrointestinal parasite communities of ring-billed gulls breeding on an island in the St. Lawrence River by using the three complementary dietary descriptors mentioned above and to investigate the link between diet and helminth communities. Each of these dietary descriptors has a distinct time frame: stomach content analysis offered evidence on recent feeding behavior. Information relating foraging and habitat-use strategy during a three-day window of time was obtained from bird-borne GPS-tracking devices. Finally, liver $\delta^{13}C$ and $\delta^{15}N$ stable isotope were analyzed offering insight into longer term food assimilation during the preceding week (Hobson and Clark, 1992). We predicted that *i*) individuals feeding more in urban habitats would have a lower abundance and diversity of gastrointestinal parasites and *ii*) individuals with similar foraging habits, as determined by the three dietary descriptors, would have similar parasite communities.

2.4 Materials and methods

2.4.1 Field sampling

Seventeen male and 17 female ring-billed gulls were collected between April 25th and June 6th, 2011 from Deslauriers Island (45°42'45''N, 73°26'25''W), a nesting colony three km northeast of Montreal (QC, Canada). This island is one of the largest ring-billed gull colonies in the St. Lawrence River, housing approximately 50 000 breeding pairs during the breeding season (May to July). Field collections overlapped only with the egg incubation period. Nests with complete clutches (i.e. three eggs) were randomly selected from all sections of the colony and one of the two nest attendants was captured using a remote controlled nest trap (Bustnes *et al.*, 2001). All birds were marked using a U.S. Fish and Wildlife Service steel ring and a colour-coded plastic band, and morphometric measurements (tarsus, culmen, bill, head length, and wing chord) as well as body mass were recorded. A miniature GPS data logger (model GiPSy2; TechnoSmArt, Guidonia, Rome, Italy) was mounted on three central rectrices using waterproof tape. The GPS units weighed 12-16 g which consisted of less than 3% of the bird body mass (mean ± SD: 483 ± 44 g). The position and speed of the gulls were recorded every 4 minutes (spatial precision <5 m) for 30.7-72.9 hours, a duration limited by the batteries of the data loggers. Recaptured gulls were euthanized by cervical dislocation and necropsied immediately on site. The mean number of days elapsed between clutch initiation and euthanasia was 22 ± 6 days (range: 8-36 days). The entire gastrointestinal tract (oesophagus to the cloaca) and liver were removed, transferred into a plastic bag and transported on ice to the laboratory, where stomach contents (i.e., oesophagus and proventriculus contents) were sorted and categorized as earthworms, insect larvae, adult insects, corn grain, pebbles, anthropogenic indigestible foods, anthropogenic foods, and plant material, and recorded as present or absent; and the intestines were surveyed for parasites. The liver was transferred into a -20°C freezer for stable isotope analyses. Capture and handling methods of ring-billed gulls were approved by the Institutional Animal Care Committee of UQAM (Montreal, QC, Canada) (permits no. 646 and 768), which comply with the Canadian Council on Animal Care (CCAC) guidelines.

2.4.2 Parasite identification

All dissections and identifications were carried out by the same individual. Twenty one of the 34 ring-billed gull gastrointestinal tracts were processed within 24 hrs of capture and the remainder (n = 13) were frozen at -80°C for subsequent examination. The gastrointestinal tracts were cut longitudinally and agitated in tap water to dislodge parasites, and the inner surface of the intestine was inspected using a stereomicroscope. Live nematodes were fixed in steaming 70% alcohol and preserved in a solution of 70% ethanol and 5% glycerol, while cestodes and digeneans were directly preserved in 95% ethanol for further identification. Specimens were stained with acetocarmine (6-8 drops/30 mL distilled water) and mounted on slides using fast drying Eukitt (45% acrylic resin and 55% xylene, EMS, Pennsylvania, USA). Roundworms were cleared in glycerine alcohol (70% ethanol/5% glycerol) and were temporarily mounted on slides with glycerol for observation and identification. The specimens were observed under a compound microscope and identified to genus, as members of the same genus have similar intermediate hosts during their life cycle, using keys from Khalil, Jones and Bray (1994) and Schmidt (1970) for cestodes; Schell (1970), McDonald (1981), Gibson, Jones and Bray (2002), Jones, Bray and Gibson (2005) and Bray, Gibson and Jones (2008) for digeneans; Anderson, Chabaud and Willmott (2009) and Wong (1990) for nematodes; and using descriptions in the literature. Abundance and prevalence of genera then were calculated according to Bush *et al.* (1997).

2.4.3 GPS data treatment

The data loggers recorded the position, speed and altitude of the GPS-tracked ring-billed gulls. ArcGIS software (ESRI, Redlands, CA, USA) was used to create a map of the Greater Montreal Area which encompassed the entire range of ring-billed gulls nesting on Deslauriers Island. GPS positions of birds in flight, that is, at altitudes above ground level and moving at speeds greater than 4 km/h (Shamoun-Baranes *et al.*, 2011) were not included in the final dataset (Patenaude-Monette, 2011). During the breeding season, gulls spend most of their time at the colony during which time they have not been observed foraging (Patenaude-Monette, 2011). Therefore, these GPS positions were removed from the final dataset as well.

All trips outside the colony were thus assumed to be related to foraging activities (Racine *et al.*, 2012). Based on the different intermediate hosts found in particular habitats used by parasites identified in ring-billed gulls, four main foraging areas were defined on the map: 1) anthropogenic areas (including urban areas and landfills), 2) freshwater areas (including lakes and rivers), 3) agricultural areas, and 4) wastewater treatment plant basins. Because duration of the GPS data-logger battery somewhat varied, creating variations in the total number of hours recorded by each unit (range: 30.7-72.9 hours), GPS data were normalized to represent the percentage of time spent in each of these four foraging habitats.

2.4.4 Stable isotope analyses

Liver stable carbon (^{13}C) and nitrogen (^{15}N) isotope analyses were carried out at the Centre de recherche en géochimie et géodynamique (GEOTOP), Université du Québec à Montréal (Montreal, QC, Canada). An aliquot of liver sample was freeze-dried (Freezone 12, Labconco, Kansas City, Missouri, USA) and ground into a homogeneous powder which were transferred into tin cups and weighed (± 0.001 mg). Isotopic measurements were carried out using a continuous flow isotope ratio mass spectrometer (Micromass Isoprime, Cheadle, UK) and an elementary analyzer (Carlo Erba NC1500, Milan, Italy). Results were reported in delta notation (δ) relative to international standards for $\delta^{13}C$ (Vienna PeeDee Belemnite (VPDB)) and $\delta^{15}N$ (air (AIR)) in per mil (‰) using the following equation: $\delta X = [(R_{sample}/R_{standard})-1]*1000$, where X was either ^{15}N or ^{13}C, and R denoted the ratio of ^{13}C:^{12}C or ^{15}N:^{14}N.

2.4.5 Conversion of trophically-transmitted parasites to implicated prey

Parasite life cycles were used to identify implicated prey (intermediate hosts). Most prey were invertebrates, in which infection levels remain generally low (Marcogliese, 1995; Locke *et al.*, 2013). Thus, it was assumed that a single parasite corresponded to the consumption of a single prey item, with the exception of *Diplostomum* spp., which is transmitted by freshwater fish. Records show that fish in the St. Lawrence River are infected by an average of five *Diplostomum* (Marcogliese and Compagna, 1999; Marcogliese *et al.*, 2006; Marcogliese *et al.*, 2010; Krause, McLaughlin and Marcogliese, 2010), and it was therefore

assumed that one fish was consumed for every five *Diplostomum* observed in bird intestines, and that fewer than five *Diplosotmum* implicated the consumption of one fish.

2.4.6 Statistical analyses

The resemblance of parasite communities, and the prey implied by parasites, was calculated between each bird using the Bray-Curtis coefficient of similarity (Clarke and Gorley, 2006). Euclidean distances of dissimilarity were derived from percentage of time spent in the different habitats and normalized stable isotope ratios of $\delta^{13}C$ and $\delta^{15}N$. Among the 34 GI tract samples, only 18 had stomach contents that contained identifiable food items, therefore, to include birds without stomach contents and which are necessarily completely similar, a dummy food item was used when deriving the Sørensen similarity indices between samples of stomach contents (Clarke, Somerfield and Chapman, 2006).

Analysis of similarity (ANOSIM), a permutation based test analogous to ANOVA, was used to test for differences in parasite communities between fresh and frozen GI tracts, as freezing may affect morphological structures used in identification of parasites. Under the null hypothesis that there is no difference between the groups of samples, *rho* would be ~ 0. No significant difference was found; therefore parasites identified from frozen and fresh carcasses were not distinguished in subsequent analysis. Differences between ring-billed gull males and females in gastrointestinal parasite communities, implicated prey, time spent in the foraging areas, stomach contents and liver $\delta^{13}C$ and $\delta^{15}N$ also were tested using ANOSIM (999 permutations). Where there were sex-specific differences, similarity percentage (SIMPER) was used to identify the variables primarily responsible for the discrimination between the groups (Clarke and Gorley, 2006). An index of size of ring-billed gulls was obtained using principal components analysis (i.e. PC1) based on three morphometric measurements: tarsus and culmen length, and wing chord. Body condition then was calculated as the residuals of the regression between body mass and PC1 for each individual.

To test whether gulls relying less on natural prey would have fewer parasites (abundance and richness), Linear and Spearman's rank correlations were performed to investigate the relationships between total parasite abundance, the number of parasite genera, Julian date of recapture, liver $\delta^{13}C$ and $\delta^{15}N$, body mass, body condition, and the percentage of time spent

in different foraging areas (agricultural, anthropogenic, freshwater and wastewater). In order to determine which of the stomach content, foraging area and stable isotope independent variables best explained the variations in the parasite communities (dependent variable), the BEST routine that identifies best rank correlations of diet descriptor variable combinations with parasite communities was used (Clarke and Gorley, 2006). To test whether birds that were similar in terms of one diet descriptor and their parasite communities, the RELATE routine (a comparative Mantel-type test which uses Spearman's rank correlation *rho* on similarity matrices of each variable) was performed with 9999 permutations. For all tests, statistical significance was assumed at α=0.05 and means are presented ± standard deviation (SD). Statistical analyses were accomplished using PRIMER 6 (PRIMER-E Ltd, Lutton, Ivybridge, United Kingdom) and the statistical package JMP (SAS, Cary, NC, USA).

2.5 Results

2.5.1 Parasite abundance and diversity

All 34 ring-billed gulls were infected with 2 – 141 parasites (47 ± 36 / individual) (Table 1). The parasites belonged to 20 genera including eight cestodes, eight digeneans and four nematodes, and implicating up to seven categories of prey (Table 2.1). Ring-billed gulls were infected by an average of 4.1 ± 1.5 genera (range: 2-9) (Table 2.2). The three most prevalent and abundant parasites were *Wardium* spp., *Diplostomum* spp. and *Eucoleus* sp. (Table 2.1). Six of the 20 parasites occurred only in single birds (*Paricterotaenia* sp., *Microsomocanthus* sp., *Plagiorchis* sp., *Apophallus* sp., *Leucochloridium* sp. and *Cardiocephalus* sp.). In order of prevalence, the prey implicated by parasites were crustaceans > freshwater fish > earthworms > insects > amphibians > marine fish and snails (Table 2.1).

Date of recapture was unrelated to total parasite abundance (Spearman's *rho* = -0.08, p = 0.65) but gulls caught at later dates had fewer *Eucoleus* sp. (Spearman's *rho* = -0.37, p = 0.03). There was no significant difference in the total abundance (Wilcoxon (rank sum) test, H = 1.76, 1df, p = 0.18), the number of parasites (Wilcoxon (rank sum) test, H = 0.50, 1df, p = 0.48) or overall parasite community structure (ANOSIM, R = 0.01, p = 0.28) between males and females. Body mass had no effect on parasite abundance (Spearman's *rho* = -0.09, p = 0.62) or number of parasites ($F_{1,33}$ = 0.25, p = 0.90). Body condition was obtained from the regression between mass and the index of structural size obtained from the PC1 of morphometric measurements of males (proportion of variation explained by component = 52.1%) and females (proportion of variation explained by component = 52.6%) separately. It also had not effect on parasite abundance (Spearman's *rho* = 0.19, p = 0.28) or the number of parasites ($F_{1,33}$ = 0.17, p = 0.69).

Table 2.1 Prevalence, mean (± SD) abundance, intensity range and implicated prey of trophically-transmitted parasites observed in ring-billed gulls (*Larus delawarensis*) ($n = 34$) collected in the Greater Montreal Area (QC, Canada)

	Prevalence (%)	Mean abundance ±SD	Intensity range	Implicated prey	Reference
Cestodes					
Wardium spp.	94	24.2 ± 25.6	1-90	Crustaceans	McDonald, 1969
Choanotaenia spp.	18	1.2 ± 5.2	1-30	Insects	McDonald, 1969
Anomotaenia spp.	12	1.0 ± 4.7	1-27	Crustaceans	Jarecka, Bance and Burt, 1984
Dilepis spp.	12	0.1 ± 0.4	1-2	Earthworms	Rysavy, 1979
Drepanidotaenia spp.	9	0.2 ± 0.9	1-5	Crustaceans	McDonald, 1969
Tetrabothrius spp.	6	0.5 ± 2.9	1-17	Marine fish	Khalil, Jones and Bray, 1994
Paricterotaenia sp.	3	0.1 ± 0.3	2	Earthworms	Gabrion and Helluy, 1982
Microsomacanthus sp.	3	0.0 ± 0.2	1	Crustaceans	McDonald, 1969
Digeneans					
Diplostomum spp.	85	9.7 ± 12.1	1-50	Freshwater fish	Palmieri, Heckman and Evans, 1976
Mesoophorodiplostomum spp.	23	4.0 ± 18.1*	2-106	Freshwater fish	Krull, 1934
Stephanoprora spp.	15	0.7 ± 3.0	1-17	Freshwater fish	McDonald, 1969
Echinostoma sp.	9	0.3 ± 1.0	2-5	Amphibians	Johnson and Mackenzie, 2008
Plagiorchis sp.	3	0.4 ± 2.6	15	Insects	McDonald, 1969
Apophallus sp.	3	0.1 ± 0.3	2	Freshwater fish	Kent *et al.*, 2004
Leucochloridium sp.	3	0.1 ± 0.3	2	Snails	McDonald, 1969
Cardiocephalus sp.	3	0.03 ± 0.2	1	Marine fish	Sudarikov and Karmanova, 1973
Nematodes					
Eucoleus (Capillaria) sp.	68	2.8 ± 3.4	1-13	Earthworms	Davis *et al.*, 1971
Cosmocephalus sp.	32	0.8 ± 2.1	1-11	Crustaceans	Wong and Anderson, 1982
Paracuaria sp.	9	0.2 ± 0.8	1-4	Crustaceans	Anderson and Wong, 1982
Tetrameres spp.	6	0.1 ± 0.2	1	Crustaceans	Quentin and Barre, 1976

*The mean abundance of *Mesoophorodimplostomum* spp. strongly reflects the one bird infected with 106 of the 136 observed in 8 of the birds sampled

2.5.2 Relationship between stomach contents and parasite communities

In the 18 samples with identifiable stomach contents (from 11 females and 7 males), major isolated food items in order of frequency included various plant parts > adult insects > earthworms > corn grain > anthropogenic nonedible food (plastic and glass) > insect larvae and anthropogenic food (rice) (Table A.I in Appendix). No difference was observed in stomach content composition between female and male ring-billed gulls (ANOSIM, R = -0.03, $p = 0.76$). No relationship was found between stomach content and parasite community or implicated prey (Table 2.3).

2.5.3 Relationship between foraging areas and parasite communities

There was no correlation between percent time spent in anthropogenic habitats and parasite abundance (Spearman's *rho* = -0.18, $p = 0.31$) or the number of parasites ($F_{1,33} = 1.46$, $p = 0.24$). However, birds that allocated foraging time among habitat types in a similar manner showed a marginal tendency to have similar parasite communities (RELATE, *rho* = 0.16, $p = 0.07$). Males with similar foraging area usage also had similar parasite communities (RELATE, *rho* = 0.34, $p = 0.01$) (Table 2.3).

Ring-billed gulls spent between 1.6 and 39.7% of their time outside of the colony (mean: 19.2 ± 10.0%). Females spent almost twice as much time outside of the colony as males ($mean_{females}$: 24.8 ± 9.3%, $mean_{males}$: 13.6 ± 7.2%; $F_{1,33} = 15.38$, $p = 0.0004$). Among GPS-tracked birds, 100% foraged in freshwater, 94% in agricultural lands, 79% in anthropogenic areas and 18% wastewater treatment basins, at any given moment. Most gulls spent the majority of their foraging time in agricultural areas (total mean: 69.1 ± 25.7%), except for one female which foraged solely in freshwater (Table 2.2). Overall, results from the ANOSIM routine showed a weak but significant differences in foraging area usage between females and males (ANOSIM, R= 0.07, $p = 0.04$). Most of this difference (SIMPER analysis, 80%) resulted from the tendency of females to spend a larger proportion of foraging time in agricultural habitats, and less time in urban habitats, than males (Table 2.2).

Table 2.2 Mean (± SD) body mass, percentage of time spent in different foraging areas, liver $\delta^{13}C$ and $\delta^{15}N$ and parasite abundance and richness observed in males (n = 17), females (n = 17) and combined ring-billed gulls (n = 34)

	Females	Males	All gulls	Range
Body mass (g)	439 ± 24	507 ± 31	483 ± 44	400-581
Time spent in different foraging areas (%)				
Agriculture	74.3 ± 25.7	63.9 ± 25.4	69.1 ± 22.8	0-97.4
Anthropogenic	10.9 ± 15.1	25.5 ± 25.3	18.2 ± 21.76	0-87.4
Freshwater	14.4 ± 23.1	8.6 ± 8.9	11.5 ± 17.6	1.1-100
Wastewater	0.4 ± 0.9	2.0 ± 5.9	1.2 ± 4.3	0-21.7
Stable isotopes (‰)				
$\delta^{13}C$	-22.5 ± 1.2	-22.1 ± 1.3	-22.3 ± 1.2	-25.1--19.1
$\delta^{15}N$	9.8 ± 0.5	9.6 ± 0.6	9.7 ± 0.5	8.8-11.0
Parasites				
Total abundance	52.8 ± 35.3	40.3 ± 36.0	46.6 ± 35.7	2-141
No. of parasite genera	4.4 ± 1.7	3.9 ± 1.3	4.1 ± 1.5	2-9

2.5.4 Relationship between stable isotopes and parasite communities

In the liver of ring-billed gulls, $\delta^{13}C$ ranged from -25.1 to -19.1‰, and $\delta^{15}N$ from 8.8 to 11‰ (Table 2.2). Stable isotope signatures did not differ between males and females (ANOSIM, R = 0.002, $p = 0.36$).

Parasites were more abundant in birds with higher $\delta^{15}N$ ($F_{1,33} = 5.05$, $p = 0.03$) (Fig. 2.1) but unrelated to $\delta^{13}C$ (Spearman's *rho* = -0.25, $p = 0.15$). There were fewer types of parasite in birds with higher $\delta^{13}C$ (Spearman's *rho* = -0.45, $p = 0.008$) (Fig. 2.2), although parasite diversity varied randomly with respect to $\delta^{15}N$ ($F_{1,33} = 0.25$ $p = 0.62$). Birds with similar stable isotopes had similar parasite communities, as evidenced by both BEST and RELATE analyses (Table 2.3). Variation in parasite community resemblance was best explained by both $\delta^{15}N$ and $\delta^{13}C$ (BEST, $r^2 = 0.34$), while other variables made no significant contribution. Birds with similar parasite communities had similar $\delta^{15}N$ and $\delta^{13}C$, regardless of whether they were partitioned by sex (RELATE, $0.47 > rho > 0.24$, $p \leq 0.05$) (Table 2.3).

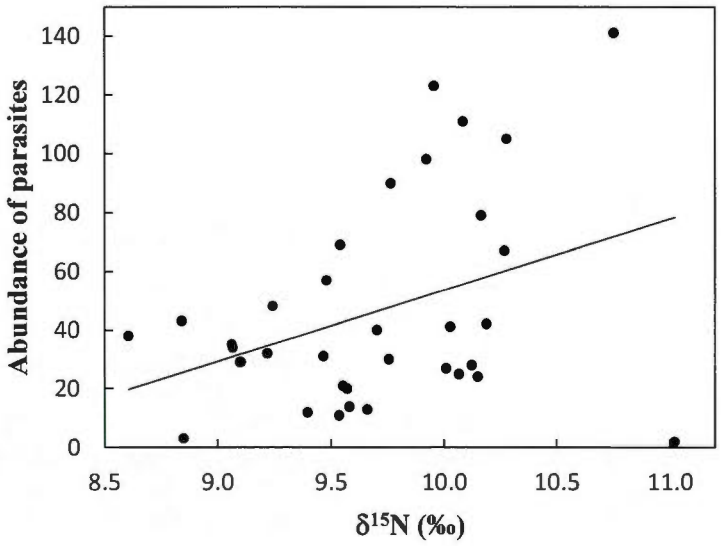

Figure 2.1 Total gastrointestinal parasite abundance plotted against liver $\delta^{15}N$ ($F_{1,33} = 5.05$, $p = 0.03$) of 34 ring-billed gulls (*Larus delawarensis*) collected in the Greater Montreal Area (QC, Canada)

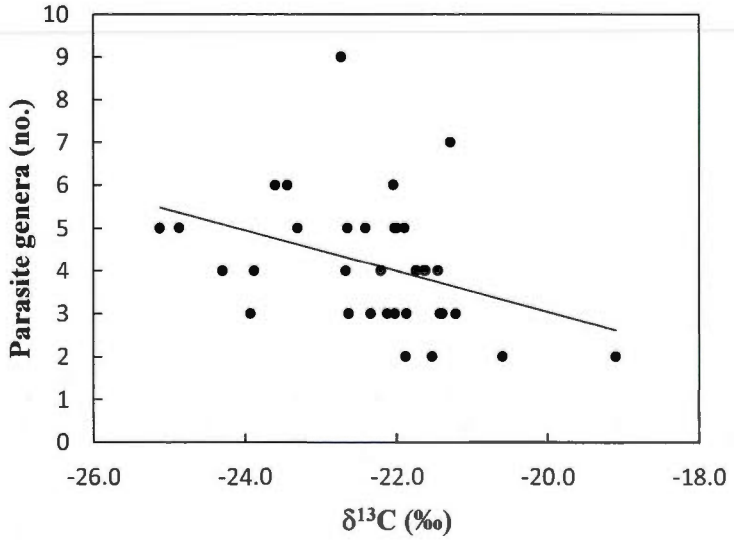

Figure 2.2 Number of genera of gastrointestinal parasites plotted against liver $\delta^{13}C$ (*rho* = -0.46, $p = 0.008$) in 34 ring-billed gulls (*Larus delawarensis*) collected in the Greater Montreal Area (QC, Canada)

2.5.5 Relationship between diet descriptors and implicated prey

Results from the RELATE routine showed that, in male ring-billed gulls, implicated prey were correlated to the foraging areas used (RELATE, *rho* = 0.36, $p = 0.008$) (Table 2.3), but not with liver $\delta^{15}N$ and $\delta^{13}C$ (RELATE, *rho* = 0.24, $p = 0.07$). Conversely, in females, a correlation was found between implicated prey and $\delta^{15}N$ and $\delta^{13}C$ (RELATE, *rho* = 0.47, $p = 0.004$), although no association was found with foraging areas (RELATE, *rho* = 0.15, $p = 0.20$). When the female and male samples were analyzed together, the significance between relationships changed, that is, implicated prey were correlated both with the foraging areas used by birds (RELATE, *rho* = 0.23, $p = 0.02$) and liver $\delta^{15}N$ and $\delta^{13}C$ (RELATE, *rho* = 0.41, $p = 0.001$) (Table 2.3).

Table 2.3 Results of (A) BEST routine (B) RELATE (a comparative Mantel-type test which uses Spearman's rank correlation *rho* on similarity matrices of each variable) routine among males (n = 17), females (n = 17) and all (n = 34) ring-billed gulls (*Larus delawarensis*) collected in the Greater Montreal Area (QC, Canada)

(A) *BEST*

Combination of variables	$\delta^{13}C$ $\delta^{15}N$	Correlation (r)	0.336

(B) *RELATE*

	Males		Females		All	
Variables	*rho*	*p*-value	*rho*	*p*-value	*rho*	*p*-value
Parasite communities and stomach contents	-0.05	0.61	-0.15	0.90	-0.11	0.91
Parasite communities and foraging areas	**0.34**	**0.01**	0.06	0.33	0.16	0.07
Parasite communities and stable isotopes	**0.24**	**0.05**	**0.47**	**0.004**	**0.38**	**0.0003**
Implicated prey and stomach contents	-0.17	0.89	-0.20	0.97	-0.16	0.99
Implicated prey and foraging areas	**0.36**	**0.008**	0.15	0.20	**0.23**	**0.02**
Implicated prey and stable isotopes	0.24	0.07	**0.47**	**0.004**	**0.41**	**0.001**

2.6 Discussion

2.6.1 Diet of ring-billed gulls inferred from parasites

In contrast to other studies, sex (e.g. Robinson *et al.*, 2010) and body mass of birds (e.g. Gregory, Keymer and Harvey, 1991) had no effect on the overall parasite community structure and composition in GI tract of ring-billed gulls, nor did their body condition. In general, birds tended to have relatively low diversity of parasites. The three most prevalent and abundant genera identified in ring-billed gulls were *Wardium* spp., *Diplostomum* spp., and *Eucoleus* sp. In a parasite survey of various fish species collected along the St. Lawrence River, at least 12 different species were found to be infected by *Diplostomum* spp. (Marcogliese and Campagna, 1999). The high abundance and prevalence of *Wardium* spp. and *Diplostomum* spp. may be an indication that ring-billed gulls consumed crustaceans (more specifically, copepods and ostracods which are the intermediate host of *Wardium* spp.) and freshwater fish more than other prey items which would necessarily harbor different species of parasites in the aquatic ecosystem. Moreover, *Eucoleus* sp., a nematode typically found in earthworms (Davis *et al.*, 1971), was likely acquired in agricultural or grassy areas (Haymes and Blokpoel, 1978; Welham, 1987). Earthworms, which are thought to be an essential source of protein (Edwards, Arancon and Sherman, 2010) for egg production, become available late in spring when the soil has thawed and coinciding with the egg-laying period (J.-F. Giroux, personal communication, 2013). The significant negative correlation between sampling date and the abundance of *Eucoleus* sp. suggests that birds reduce their consumption of earthworms as the incubation period progresses. This seems to overlap with the period of time when crops (e.g. soya and corn) begin to grow in agricultural lands, making these grounds less accessible for feeding on earthworms (Patenaude-Monette, 2011). Interestingly, *Paricterotaenia* sp., also found in earthworms, were not always found in birds which were infected by *Eucoleus* sp. This would suggest that *Paricterotaenia* spp. may not be common in the Greater Montreal Region.

2.6.2 Relationship between parasites and a very short term diet descriptor

Each of the three different diet descriptors gave complementary information on the dietary composition of ring-billed gulls. The lack of relationship between stomach contents and parasite communities may be due to differences in temporal scales between these two variables. Stomach contents are known to only depict the overall diet of a species over a very short time-frame (Jackson, Duffy and Jenkins, 1987). Therefore, establishing a link between such a short-term diet descriptor with biases for certain food items that are slower to digest and parasites that reflect longer term feeding behaviour was more difficult to establish.

2.6.3 Relationship between parasites and an intermediate term diet descriptor

Males and females differed in their foraging strategies which had implications for the relationship between foraging areas used and parasite communities: similar foraging strategies were associated to similar parasite communities in males, but not in females. The lack of relationship between recent foraging areas used and the composition of parasite communities in females may suggest that their foraging strategies were less consistent during the breeding season, specifically in the pre-laying and incubation period, than males. These behavioural variations may be due to biological factors associated with their breeding ecology and reproductive role. In contrast, males seemed to exhibit a higher degree of foraging specialization at a local scale. Sex-specific differences associated with foraging behaviour during the incubation period have also been observed in other colonially breeding bird species (e.g. black-browned and grey-headed albatross, *Thalassarche melanophrys*, *T. chrysostoma*) (Philips *et al.*, 2004).

Although the present study was unable to establish a significant link between recent utilization of foraging areas and parasite communities for combined males and females, it should not necessarily be presumed that no such link exists. The three day time-frame used to track ring-billed gulls was at least long enough to demonstrate trends in the relationship between recent foraging behaviour and parasite communities. The broad categorization of habitats used in the study may not have been refined enough to detect the subtle variations that occur within host-parasite assemblages. Since many different parasites are transmitted

through the same types of prey, reducing the number of parasite variables by converting the parasite data to implicated prey yielded stronger relationships with foraging areas used by the birds. Birds feeding on similar implicated prey types also had similar foraging habitat use strategies as revealed by the information recorded by the GPS data loggers during the last two to three days monitored.

This study is novel for its use of GPS-based tracking information on foraging area usage strategies to explain parasite community structure. Based on the transmission pathways of gastrointestinal parasites, it can be assumed that birds feeding in anthropogenic areas such as cities and landfills are less likely to encounter parasites. However, the present study found no significant negative correlation between the time spent in anthropogenic areas and parasite abundance or diversity in ring-billed gulls. This lack of relationship may be attributed to the scale of the study (i.e. only a single colony), or by some of the basic assumptions regarding the behaviour of the birds outside of the colony. Due to the nature of our study design, it could not be determined with certitude whether the areas visited were used for foraging or for other behaviors (e.g. resting, preening, etc.). Geographical movement analysis revealed that most birds spent the majority of their time in agricultural fields, except for one female bird which spent all of its time in freshwater habitats (various lakes as well as the St. Lawrence River and its tributaries). Although, in general, birds did not spend a large percentage of time in freshwater areas relative to agricultural or anthropogenic areas, it is interesting to note that all birds visited freshwater habitats.

2.6.4 Relationship between parasites and a longer term diet descriptor

Liver $\delta^{13}C$ and $\delta^{15}N$ signature in ring-billed gulls best explained the variations observed in parasite community composition and abundance, thus suggesting that differences in foraging ecology can affect a host's exposure to parasites. More specifically, individuals with similar $\delta^{13}C$ and $\delta^{15}N$ were found to have similar parasite communities. The liver $\delta^{13}C$ and $\delta^{15}N$ provided an idea of the nutrients that were assimilated over the last four to ten days (Hobson and Clark, 1992), while parasites reflected historical feeding behaviour as they were likely acquired over a longer period of time (from weeks to months depending on the group). As a consequence, any parasite recruited over the last four days would not yet be mature.

However, most of the parasites sampled were mature adults which must have been acquired more than four days prior to sampling. Therefore, the associations between stable isotopes and parasite communities suggests that individuals within this species have a tendency to conserve particular foraging habits throughout the pre-laying and incubation period, consistent with previous studies on ring-billed gulls (Welham, 1987; Caron-Beaudoin *et al.*, submitted).

The link between stable isotope signatures and parasites has also been found in double-crested cormorants (Robinson, Forbes and Hebert, 2009). However, Robinson, Forbes and Hebert (2009) only investigated the correlation between one type of parasite (*Contracaecum* spp.) and $\delta^{13}C$ and $\delta^{15}N$. Furthermore, they found no association between $\delta^{15}N$ and the abundance of *Contracaecum* spp., whereas the present study uncovered a significant positive relationship between $\delta^{15}N$ and total parasite abundance. Caron-Beaudoin at al. (submitted) showed that the presence of human-made foods in the diet of ring-billed gulls was associated with a lower $\delta^{15}N$ value in the liver. Although the present study did not find a link between time spent in anthropogenic habitats and total parasite abundance, a link can still be inferred from $\delta^{15}N$. Lower $\delta^{15}N$ values in the liver, associated with a more human-based diet, were linked to a lower abundance of intestinal parasites. This would confirm the hypothesis that ring-billed gulls feeding on more anthropogenic sources of food would be less likely to encounter trophically-transmitted parasites. $\delta^{13}C$ is used to compare pelagic vs. littoral consumers (France, 1995) for seabird species (Hobson, Piatt and Pitocchelli, 1994). Robinson, Forbes and Hebert (2009) found a negative correlation between *Contracaecum* spp. and $\delta^{13}C$. From this, the authors concluded that double-crested cormorants feeding in more pelagic areas were more likely to be exposed to *Contraceacum* spp. nematodes. Despite the fact that a significant negative correlation was also found between total parasite abundance and $\delta^{13}C$ in our ring-billed gulls, it is not possible to make clear inferences from the $\delta^{13}C$ of this omnivorous species. Caron-Beaudoin *et al.* (submitted) reported that the presence of varying proportion C_3 and C_4 plants, from wheat (found in bread) or corn (found in many human-made foods), makes their carbon isotope signature more difficult to interpret.

2.7 Conclusion

For the first time, it was shown that the accessibility of anthropogenic sources of food has an effect on the abundance of trophically-transmitted parasites in an urban adapted avian species. In previous studies, it has been found that anthropogenic sources of food comprise an important part of the diet of ring-billed gulls. Through the use of stable isotopes, this study showed that ring billed-gulls having a more human-based diet encounter less trophically-transmitted parasites, although this could not be corroborated with the percent time they spent in anthropogenic habitats as determined from the GPS-tracking information recorded during the three day time-frame preceding sampling. The use of GPS-tracking information collected over a longer time-frame may provide better resolution for future studies. To our knowledge, this is the only study to relate information on recent foraging area utilization obtained from GPS-based tracking information to parasite communities. This is also the first study to investigate the effect of host diet, using multiple dietary descriptors, on parasite community composition in a free-ranging avian species. According to our results, liver $\delta^{13}C$ and $\delta^{15}N$ in ring-billed gulls have the greatest potential to explain the variations observed in parasite communities of an omnivorous bird breeding in a heterogeneous landscape. Moreover, parasites which are often an ignored yet integral component of ecosystems (Marcogliese 2004) can themselves be a useful tool in understanding host diet (Marcogliese & Cone 1997). Here, the temporal change in the abundance of *Eucoleus* sp. in ring-billed gulls provided evidence that the timing of the emergence of agricultural crops have an impact on their intake of earthworms.

2.8 Acknowledgements

This work was supported by the Natural Sciences and Engineering Research Council of Canada (NSERC) through a discovery grant to J.V. and a Collaborative Research Development Grant to J.-F.G. We thank Agnieszka Adamowicz and Dr. Jean-François Hélie (GEOTOP-UQAM) for their help with the stable isotope analyses. We are grateful to Francis St-Pierre and Mathieu Tremblay (UQAM) for their fieldwork assistance and Laurine Bandet for her help with the GI tract dissections.

CONCLUSION

Understanding parasite communities and the factors involved in shaping them is important to the overall understanding of food webs, as helminths are transmitted by predator-prey interactions (Marcogliese, 2004). The diet of a predator is a proximate factor involved in shaping intestinal helminth communities. Intraspecific variations in feeding behaviour within an omnivorous species may produce intraspecific variations in parasite communities. Studies which have previously investigated this subject have used descriptors of host diet such as stomach contents as well as carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) stable isotope signatures in various tissues (e.g. muscle, liver, and feathers) (Kelly, 2000). To date, no other study has used GPS-tracking information documenting foraging habitat use strategies to explain parasite communities in any species. Moreover, contrary to other parasitology studies which often only include subsamples of a host's parasite community, the present project is notable for its inclusion of the entire gastrointestinal parasite infracommunity in all of the analyses.

Over the years, human populations have grown exponentially and as a result, urbanization has had significant impacts on local wildlife populations. There is, at present, a lack of knowledge concerning the impact these landscape changes are having on parasite communities (Delgado and French, 2012). Endoparasites with complex life cycles requiring multiple hosts are particularly vulnerable to environmental changes (Marcogliese, 2004). The loss of an intermediate host within an ecosystem could ultimately result in the disappearance of a parasite species. Furthermore, omnivorous species adapted to human-dominated habitats are likely reducing their ingestion of parasites which could also lead to the inability of parasites to complete their life cycles. As a dominant species in the Greater Montreal Area, ring-billed gulls likely have an important role in the St. Lawrence ecosystem as a vector of local parasites (Marcogliese *et al.*, 2001). Changes to host-parasite interactions can have implications for ecosystems as a whole (Minchella and Scott, 1991; Marcogliese, 2004). Helminths have generally been associated with the negative pathological effects they have on their host. However, they have an essential role in ecosystem dynamics; from controlling biodiversity to having physiological and behavioural effects on individual hosts (Marcogliese, 2004).

The first objective of this Master's project was to examine the link between the diet of an omnivorous species using both terrestrial and aquatic ecosystems, the ring-billed gull, and parasite communities; and whether individuals with similar feeding behaviours have similar parasite communities. Using this urban avian host species, the second objective was to investigate the potential effect of urbanization on endoparasite assemblages. In order to accomplish this, three dietary descriptors with different time-frames were used. Stomach contents revealed very recent food consumption. GPS-tracking provided information on habitat use strategies during the last three days; and liver carbon and nitrogen stable isotopes provided insight into food assimilation over the last week (more or less) (Hobson and Clark, 1992). Using GPS locations of ring-billed gulls, precise information on their foraging habitat use strategies while outside of the colony could be obtained. It was assumed that these sites which included agricultural, anthropogenic (cities and landfills), freshwater areas and wastewater treatment basins were used for foraging. However, it is possible that these sites were also used for other activities such as resting or preening. This remains a confounding factor in this study.

The helminths found in the 34 ring-billed gulls sampled revealed that this species consumed considerable amounts of crustaceans (infected with *Wardium* spp.) (McDonald, 1969) and freshwater fish (infected with *Diplostomum* spp.) (Palmieri, Heckman and Evans, 1976) probably prior to their incubation period. Furthermore, a reduction in the abundance of *Eucoleus* sp., a nematode infecting earthworms (Davis *et al.*, 1971), as the incubation season progressed suggests that ring-billed gulls feed less on earthworms in late May and early June. This may be associated with nearby agricultural practices (Patenaude-Monette, 2011).

Stomach contents were not associated with the structure of parasite communities. However, individuals with similar foraging habitat use strategies (that is, the percentage of time spent in different foraging sites) tended to have similar parasite communities. Though, $\delta^{13}C$ and $\delta^{15}N$ ratios were found to best explain variations in parasite assemblages. While no link was found between the percentage of time spent in anthropogenic areas and total parasite abundance of individual ring-billed gulls, a link between anthropogenic food consumption and parasite loads could be inferred from $\delta^{15}N$ ratios. More specifically, individuals with lower $\delta^{15}N$ had significantly less endoparasites (total number). It has previously been shown that anthropogenic food sources tend to be depleted in ^{15}N (Jahren and Kraft, 2008; Caron-

Beaudoin *et al.*, submitted). Thus, it may be inferred that ring-billed gulls feeding lower in the food chain or similarly, feeding more on human-made foods (e.g. fast food) are ingesting fewer parasites. For future studies, it would be interesting to compare the abundance and prevalence of parasites of different populations of gulls with varying degrees of reliance on anthropogenic foods. Such a study would shed more light on the impact that urbanization can have on parasite communities. Given that parasites reflect long term feeding behaviour, GPS-tracking information acquired over a longer period of time (more than the three day time-frame used in this study) would likely reveal stronger associations between foraging habitat use strategies and parasite community structure of an omnivorous species foraging in a heterogeneous landscape.

Parasites have a number of different impacts on their host. They impose energetic demands on their host; they affect host sex ratios and mate choice; they alter host behaviour; they reduce fecundity and in some cases they cause host mortality. Consequently, parasites may impact host fitness, reproductive success and demography. Together, these effects can be important determinants in ecosystems and population dynamics of wildlife species (Minchella and Scott, 1991). The success of ring-billed gulls near urban areas between 1980 and 2000 has often been attributed to the abundance of human-made foods in cities and landfills (Brousseau, Lefebvre and Giroux, 1996; Belant, Ickes and Seamans, 1998). However, an indirect and previously overlooked consequence of the availability of anthropogenic food sources is reduced rates of exposure to trophically-transmitted parasites, consequently reducing their parasite infection levels. A decline in the overall impact of parasites on this avian host may have also contributed to their population growth during that time. However, since then, stricter regulations regarding the covering of human waste in landfills have been emplaced and are likely affecting the availability of anthropogenic food. As organisms with an important role in host population control, increases in parasite infections due to a shift back to natural sources of food (i.e. infected prey) could be contributing to the slight decline observed in the population of ring-billed gulls since 2000. A study investigating the potential historical change in parasite community structure in ring-billed gulls breeding in the St. Lawrence Region may provide answers to some of the current questions regarding the impacts of helminths on the population of this urban species.

APPENDIX

Table A.1: Presence or absence of edible and non-edible food items isolated from proventriculus and gizzard of the 18 ring-billed gulls (*Larus delawarensis*) collected in the Greater Montreal Area (QC, Canada) in which stomach contents were present.

Bird ID #	Corn grain	Earthworms	Adult insect	Insect larvae	Pebbles	Anthropogenic nonedible food	Plant parts	Anthropogenic food
CAO					✓		✓	
CTA		✓				✓		
HUU	✓							
JPA					✓		✓	
JUK	✓							
LJC			✓					
LKZ		✓				✓	✓	
MCA	✓		✓			✓		
MHM		✓					✓	
MMC							✓	
MPT	✓	✓	✓				✓	
UFF		✓	✓		✓		✓	
UFH			✓				✓	
XHL		✓	✓				✓	
XKK						✓		
ZJU		✓			✓		✓	
ZKC			✓					
ZLL		✓	✓	✓			✓	
ZLM		✓			✓		✓	
ZMP		✓	✓		✓		✓	
ZTC			✓					
ZTP	✓		✓				✓	
ZXC								✓

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